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Reclamation of Abandoned Mine Soil Using Biosurfactant

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***Abstract:** Mining results in enormous land damage, varying the ecosystem of microbial culture and disturbing plant life leading to annihilation of the land. Due to this researcher's has shifted their focus to alternative methods for the sustainable development. The present study aims at environmental friendly and cost effective technique for the reclamation of abandoned mine soil using bio-surfactant. In the current investigation bio-surfactant was produced using Bacillus Subtilis MTCC no. 1427 on 20% spent wash collected from distillery unit. The mined soil with bio-surfactant was able to minimize heavy metal concentration and the plants grown on this soil were healthy similar to the results of the agricultural soil.*
***Keywords:** Mining, Abandoned, Reclamation, Heavy Metals, Re-vegetation, Bioremediation.*

I. INTRODUCTION

Lithosphere is one of the most essential resources which are being exploited due to continuous progression in economic development, industrial expansion, rapid urbanization, population growth, advanced science and technology. Rapid growth in civilization thus greatly relies on mining industry [1]. The main sources of heavy metals which contribute in soil contamination are via foundries, smelting operations, mining the metal, metal leaching from various sources, and other milling operations which include screening, concentrating ores, grinding, removal of tailings, mine and mill waste water disposal [2]. The mining activities results in modification of land forms which comprises of loose soil piles, waste rocks, bare stripped areas, overburden surfaces and subsided land slopes.

The most general heavy metals found at contaminated sites are Arsenic (As), Cadmium (Cd), Chromium (Cr), Cobalt(Co), Copper (Cu), Ferrous (Fe), Lead (Pb), Manganese (Mn), Mercury (Hg), Nickel (Ni), Tin (Sn), Zinc (Zn), etc. Soils are greatly affected by the heavy metals that are released into the environment by aforesaid anthropogenic activities [3]. Reclamation is a process which helps to restore land productivity, establish biotic ecosystem, stable nutrient cycles and microbial processes [4] - [5].

Reclamation includes management of all types of physical, chemical and biological disturbances of soil like changes in fertility, microbial activity, soil pH and several nutrient cycle that make degraded soil fertile. Efficiency of land can be augmented by addition of natural products such as animal manure, saw dust, wood residue, sewage sludge which provides nutrients like nitrogen, phosphorus, potassium and organic carbon to the soil. Revegetation is one of the most broadly used remediation technique to reclaim abandoned mine soil. Revegetation reduces the soil erosion and protects against the degradation of top soil. The plant species selected for revegetation are focused on nitrogen fixing, metal tolerant, acid tolerant and healthy growth of plants. Bioremediation is a process which utilizes microorganism to eliminate the pollutants from the contaminated environment. Bio-surfactants are biodegradable and prepared using industrial by-product which will reduce the cost of production. Recently Jayalatha and VeenaKumaraAdi reported Restoration of Physico-Chemical Properties of Zinc Contaminated Soil by Bacterial Biosurfactant [6]

Reclamation of mine soil has become integral part sustainable development to restore the land forms and vegetation. This paper discuss on post mining use of land forms and control of soil pollution using bio-surfactant and revegetation by growing suitable plants to control heavy metals.

In this paper we discuss about four types of soil and their impact on the plant growth. The soil used for the analysis are agricultural soil, mined soil that is collected from the abandoned mine, mined soil mixed with the spent wash which is collected from distillery unit and the mined soil which is mixed with bio-surfactant.

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II. MATERIALS AND METHODS

A. Production Of Biosurfactant

The microorganism used in the present study for biosurfactant production namely *Bacillus Subtilis* MTCC no. 1427 was collected from Microbial Type Culture Collection & Gene Bank (MTCC), CSIR-INSTITUTE OF MICROBIAL TECHNOLOGY, sector 39-A, Chandigarh-160036, India. Nutrient and Luria broth was prepared in 250ml conical flask in the laboratory which had been autoclaved at 121ºC for 20min at 15psi. Dry freezed Bacteria were introduced into the flask containing Luria broth in the laminar air flow to avoid contamination and kept in the rotary shaker set at 37ºC and 110rpm for 24 hours. Later streaked on to the test tubes and petridishes containing solidified nutrient agar. A loop of Bacillus Subtilis cells were transferred into the 500ml conical flask contain 20% dilution of spent wash medium as a substrate which had been autoclaved for 20 min at 121ºC and kept in rotary shaker for 144 hours at 37ºC and set at 100 rpm. Later the cell free supernatant was composed and utilized as biosurfactant. Spent wash used for growing bacteria was collected from Samsons Distilleries Unit, Indian cane power Ltd. Duggavathi-583137, Davanagere District, Karnataka, India. Spent wash was stored at room temperature till further use.

B. Experimental Setup

Abandoned mine soil samples were collected from iron ore mines near Sandur, Ballari District, Karnataka and agricultural soil was collected from Bhujanganagara village near Sandur Taluk, Ballari District, Karnataka. The soil required for analysis was oven dried and sieved through 2mm sieve and kept at room temperature for further analysis.

Different soil conditions were maintained in the present study namely,

1) *Agricultural soil:* Natural existing soil was used for the experiment. About 3 kilograms of soil sample was used.
2) *Mined soil:* kilograms of mined soil sample was used.
3) *Mined soil with bio-surfactant:* In about 3 kilograms of soil sample, 300ml of biosurfactant was introduced and the contents were mixed well to get homogeneous mixture.
4) *Mined soil with spent wash:* In 3 kilograms of soil sample, 300ml of spent wash was mixed by considering 20% dilution factor that is 60ml of spent wash in 240 ml of water.

C. Instrumental Analysis

Instruments used for biosurfactant production and soil analysis are pH meter, conductivity meter, analytical balance, atomic absorption spectrophotometer, double distillation unit, mechanical rotary shaker, flame photometer, magnetic shaker, hot air oven, autoclave, etc.

For soil analysis standard procedure were adopted to determine the soil properties. The pH meter was used to determine the pH values of the soil according to Potentiometric method [7]. The soil filtrate is used to measure the conductivity of the soil using conductivity meter and expressed in terms of dS/m [8]. The specific gravity of the soil determined according to the test specified in [9]. Moisture content in the soil sample was determined by oven drying method as specified in [10] and the values are expressed in terms of percentage. Particle density of the soil was done using weighing bottle method and the values are represented in terms of g/cc [11]. Soil texture of the soil sample was determined by jar method and denoted in terms of percentage. Organic carbon of the soil sample was determined by titrimetric method and values are expressed in percentage [12]. The nitrogen content of the soil sample is determined by alkaline permanganate method and expressed in terms of kg/Ha[13]. Phosphorus in the soil sample was determined by Olsen's method and represented in terms of kg/Ha [14]. Potassium present in the soil is extracted using ammonium acetate and analyzed by flame photometer [15]. Sulphur present in the soil was determined by turbidimetric method with extraction using 0.15% of $CaCl_2$ solution, measured by spectrophotometer and expressed in terms of ppm. Iron, manganese, copper, zinc available in the soil was measured by atomic absorption spectrophotometer using DTPA extractant and expressed in terms of ppm. Boron present in the soil sample was determined by Azomethine-H Colorimetric Method and denoted in terms of ppm [16].

III. RESULTS AND DISCUSSION

Soil is one of the most important components in the plant growth which provides nutrients, support and flow of air, water network through the plant's root. Plants use nutrients present in the soil for their growth and hold together which prevents erosion. The heavy metal mobility depends on the metal concentration in the soil and properties like pH, electrical conductivity, organic matter, etc. The physio-chemical properties of the agricultural soil, mined soil, mined soil with spent wash and mined soil with biosurfactant were analyzed. The mined soil was collected from abandoned iron ores mines near Sandur, Ballari District, Karnataka and agricultural soil was collected from Bhujanganagara village near Sandur Taluk, Ballari District, Karnataka.

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Table 1: classification of standard limits for soil properties (University of Agricultural & Horticultural Sciences, Shivamogga - 2015)

pH of soil	conditions
Less than 5.5	Soil is acidic in nature
5.5 to 7.5	Normal range soil
More than 7.5	Soil is alkaline in nature

Electrical Conductivity (EC) (dS/m)	Soil Salinity
Less than 1.0	Adequate
1.0 to 2.0	Critical
More than 2.0	Harmful

Prime/Major Nutrients	Very Low	Low	Medium	High	Very High
Organic carbon (%)	Less than 0.25	0.25 to 0.50	0.5 to 0.75	0.75 to 1.0	More than 1
Nitrogen (kg/Ha)	Less than 140	140 to 280	280 to 560	560 to 700	More than 700
Phosphorus (kg/Ha)	Less than 5	5 to 10	10 to 25	25 to 40	More than 40
Potassium (kg/Ha)	Less than 60	60 to 120	120 to 280	280 to 560	More than 560

Available Nutrients (Ppm)	Critical Limit	Remarks
Zinc	Not less than 0.6	The values more than the critical limit indicates sufficient Available nutrients
Iron	Not less than 4.5	
Copper	Not less than 0.2	
Manganese	Not less than 2.0	
Boron	Not less than 0.5	
Sulphur	Not less than 10	

Table 2: Geotechnical Features of the Soil Samples

Sl.No	Parameters	Specific Gravity	Moisture Content %	Particle Density G/Cc	Soil Texture		
					Sand %	Silt %	Clay %
1	Agricultural Soil	2.21	19.1	1.2	12.5	63.8	23.6
2	Mined soil	2.8	2.2	1.5	-	82.2	17.7
3	Mined Soil + biosurfactant	2.8	2.2	1.5	-	82.2	17.7
4	Mined Soil + Spent wash	2.8	2.2	1.5	-	82.2	17.7

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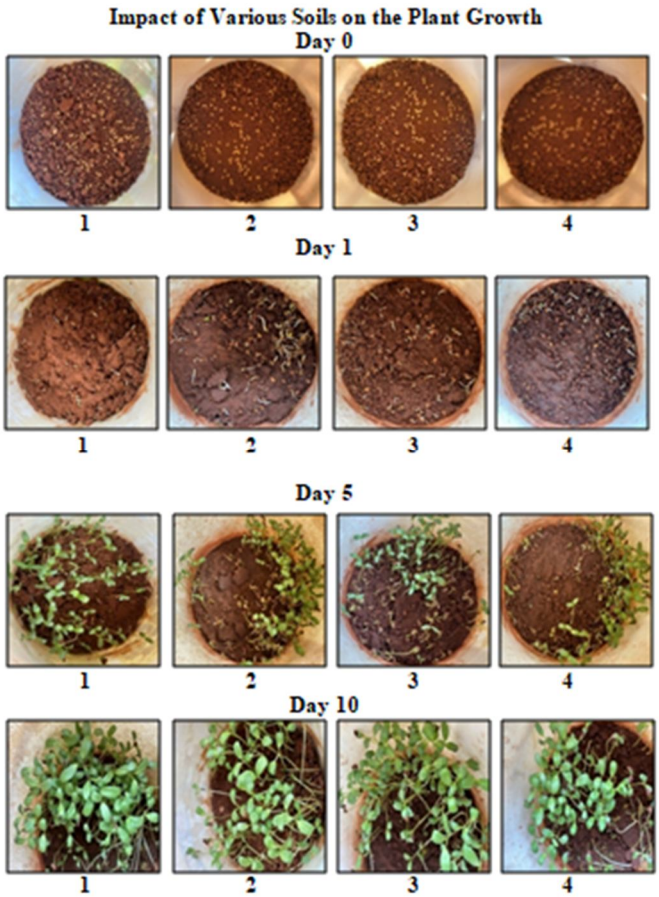
Table 3: Major and Micro Nutrients of the Soil Samples

Sl.No	Parameters	Agricultural Soil	Mined Soil	Mined Soil + Bio-Surfactant	Mined Soil + Spent Wash
1	pH	7.3	8.3	7.1	6.7
2	Electrical Conductivity (dS/m)	0.08	0.3	0.38	0.41
3	Organic Carbon (%)	1.2	0.5	0.3	0.3
4	Nitrogen (kg/Ha)	-	-	-	-
5	Phosphorus (kg/Ha)	109	117.6	33.13	33.28
6	Potassium (kg/Ha)	137	98.5	421.7	453.4
7	Sulphur (ppm)	59	80.61	10.81	61.20
8	Zinc (ppm)	3.1	19.61	0.46	0.16
9	Boron (ppm)	0.5	1.1	0.6	0.3
10	Iron (ppm)	13.5	9.97	2.19	2.21
11	Manganese (ppm)	6	9.77	4.68	4.21
12	Copper (ppm)	1.7	2.90	1.35	0.95



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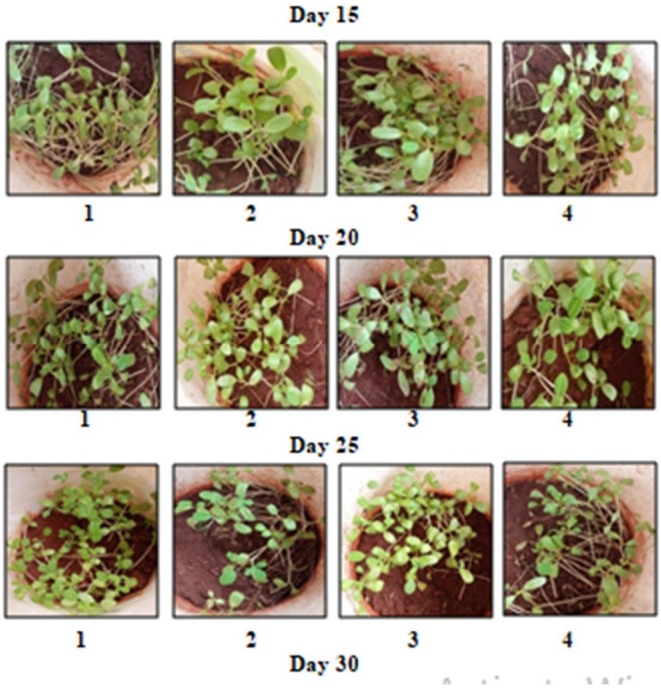

Figure 1: plants growth in the 30 days period where 1) agricultural soil, 2) mined soil, 3) mined soil with biosurfactant and 4) mined soil with spent wash.

Final Growth Result

1 2 3 4

Figure 2: comparing healthy and dead plants on the final growth where 1) agricultural soil, 2) mined soil, 3) mined soil with biosurfactant and 4) mined soil with spent wash.

From the above image we can observe that application of seeds directly to the mined soil has resulted in the poor growth of plants where dead plants are more than the healthy ones. In case of mined soil mixed with the spent wash have slight good results than the only mined soil but not as good as agricultural soil and mined soil with bio-surfactant. It was observed that mined soil with bio-surfactant has complimentary results and has almost good yield as agricultural soil. From the present study we can note that abandoned mined soil can be reclaimed by best use of mined soil in bio-surfactant. There are several studies on the reclamation of abandoned mine soil using bio-surfactant, some of the reviews and articles in [17] - [19] are related to the present study.

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IV. CONCLUSION

Heavy metals concentration has become one of the major problem in the abandoned mine sites. There are many physical, chemical and biological remediation technologies available for the reclamation of abandoned mine site. One such technique is bioremediation which is environmental friendly and cost effective method. The results are shown complimentary where the values of mined soil were compared to the mined soil with bio-surfactant. The pH value was reduced from 8.3 to 7.1 that are from alkaline to normal range. Organic carbon was reduced from 0.5 to 0.3%, Phosphorus was reduced from 117.6 to 33.13kg/Ha. Potassium was increased from 98.5 low range to 421.7kg/Ha high range. The heavy metals were optimized such as iron was reduced from 9.97 to 2.19ppm, zinc was reduced from 19.61 to 0.46ppm, boron was reduced from 1.1 to 0.6ppm, sulphur was reduced from 80.61 to 10.81ppm, manganese was reduced from 9.77 to 4.68ppm and copper was reduced from 2.90 to 1.35ppm. Similarly the results were compared with the mined soil with spent wash and agricultural soil. In the current study, results of plants grown were complimentary in the agricultural soil and in the mined soil mixed with bio-surfactant. It was observed that the mined soil having bio-surfactant was having better results than the mined soil with only spent wash. Application of seeds directly to the mined soil resulted in poor growth of the plant. From the present study we conclude that abandoned mine soil can be reclaimed by using bio-surfactant.

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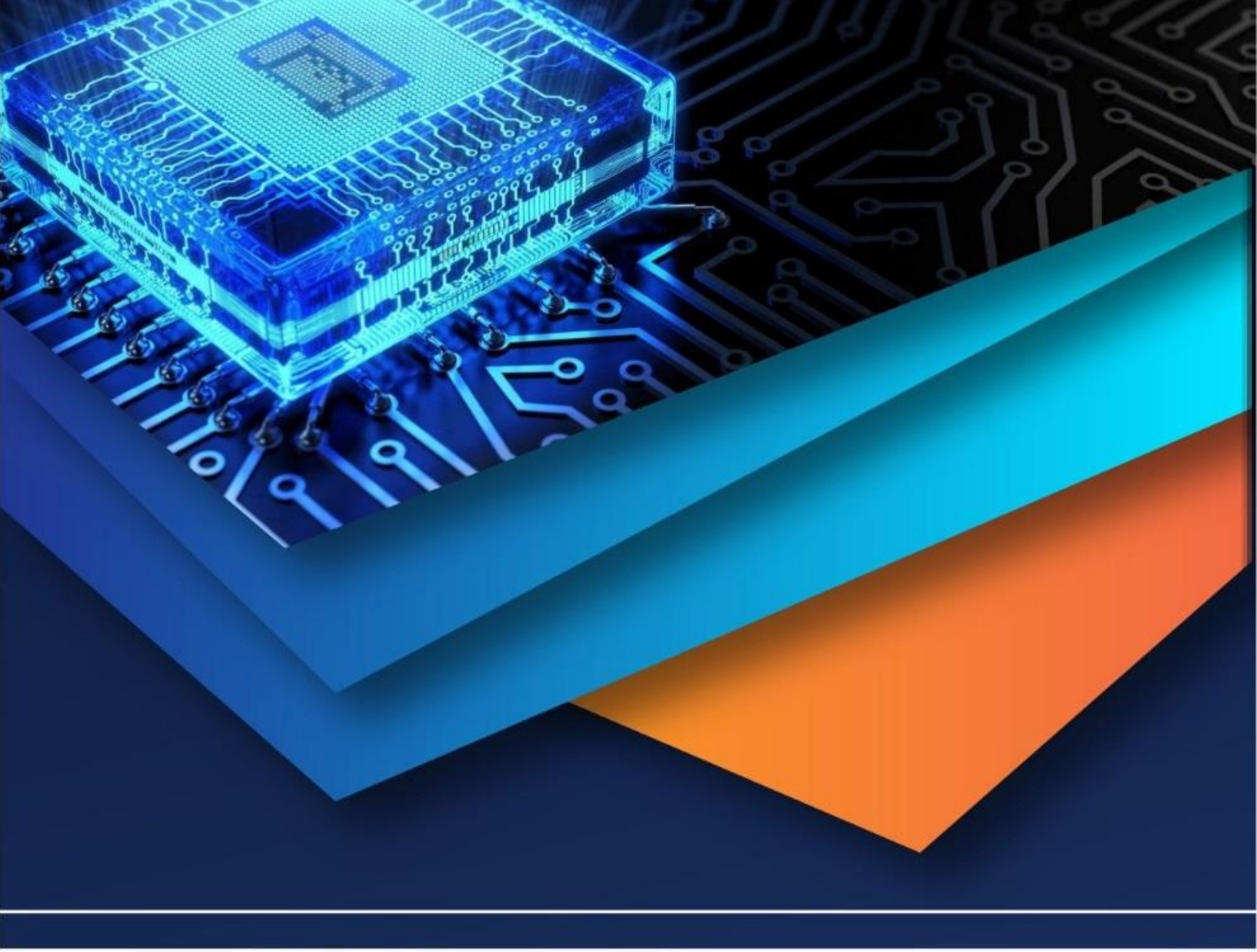

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